



Baker's Asthma: Is the Ratio of Rye Flour-Specific IgE to Total IgE More Suitable to Predict the Outcome of Challenge Test Than Specific IgE Alone

V. van Kampen, I. Sander, R. Merget, T. Brüning, and M. Raulf

Abstract

Usually the diagnosis of baker's asthma is based on specific inhalation challenge with flours. To a certain extent the concentration of specific IgE to flour predicts the outcome of challenge test in bakers. The aim of this study was to evaluate whether the ratio of specific IgE (sIgE) to total IgE (tIgE) improves challenge test prediction in comparison to sIgE alone. Ninety-five bakers with work-related respiratory symptoms were challenged with rye flour. Total IgE, sIgE, and the sIgE/tIgE ratio were determined. Receiver operator characteristic (ROC) plots including the area under the curve (AUC) were calculated using the challenge test as gold-standard. Total IgE and sIgE concentrations, and their ratio were significantly higher in bakers with a positive challenge test than in those with a negative one ($p < 0.0001$, $p < 0.0001$, and $p = 0.023$, respectively). In ROC analysis, AUC was 0.83 for sIgE alone, 0.79 for tIgE, and 0.64 for the ratio. At optimal cut-offs, tIgE, sIgE, and the ratio reached a positive predicted value (PPV) of 95%, 84% and 77%, respectively. In

conclusion, calculating the ratio of rye flour-sIgE to tIgE failed to improve the challenge test prediction in our study group.

Keywords

Allergy · Baker's asthma · Immunoglobulin E · Inhalation challenge · Occupational allergy · Rye flour

1 Introduction

Baker's asthma is one of the most frequent forms of occupational immunoglobulin E (IgE)-mediated allergy. In 2014, 64% of 584 confirmed cases of occupational asthma in Germany were caused by bakery-derived allergens, especially wheat and rye flour (Deutsche Gesetzliche Unfallversicherung (DGUV 2015)). In general, but especially within the scope of compensation claims, the specific inhalation challenge with suspected occupational allergens is considered the gold standard for the diagnosis of occupational asthma (Vandenplas et al. 2017; Muñoz et al. 2014). Since the specific challenge test is cumbersome, has a potential for severe adverse effects, and should only be performed at specialized centers, alternative methods to diagnose flour allergy are of great value.

The general association between the clinical responsiveness to allergens and the results of

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specific IgE (sIgE) tests is well known. Especially for food allergens (Buslau et al. 2014; Soares-Weiser et al. 2014) but also for ubiquitous inhalation allergens (Douglas et al. 2007; Fernández et al. 2007) it was shown that the sIgE determination can be useful to predict the result of an oral or bronchial challenge. Concerning inhalative occupational allergens, it has been demonstrated for latex and flours that high levels of sIgE have a high positive predictive value (PPV) for occupational asthma (Vandenplas et al. 2016; van Kampen et al. 2008).

The level of tIgE, which is the sum of all sIgE, is mostly elevated in case of allergic sensitization. Thus, a given sIgE level might be associated with the total IgE level, which is a potential indicator of atopy (Ahmad Al Obaidi et al. 2008). For this reason, in a couple of studies the ratio of allergen-sIgE to tIgE has been used to predict the outcome of food challenge. While some authors have found a benefit using the sIgE/tIgE ratio compared with sIgE alone (Horimukai et al. 2015; Gupta et al. 2014), others have reported the opposite (Grabenhenrich et al. 2016; Mehl et al. 2005).

Therefore, this study seeks to define the diagnostic utility of the ratio of rye flour-sIgE to tIgE in the diagnosis of baker's asthma. Due to the fact that tIgE levels are mostly elevated in atopic subjects, data were additionally stratified according to atopy status.

2 Methods

2.1 Study Design

The study was approved by the Ethic Committee of the Ruhr University Bochum in Germany, and all study participants gave informed written consent. Ninety-five bakers (mean age of 40 ± 13 years, range of 19–76 years, 79% of males) with work-related symptoms of asthma and/or rhinitis were included in the study. All subjects answered a questionnaire and underwent a clinical examination. In addition, tIgE and rye flour-sIgE determinations, as well as a challenge test with rye flour were performed. All the bakers

were examined within the scope of claims for compensation due to occupational asthma.

2.2 IgE Determination

Total IgE and rye flour-sIgE were measured by ImmunoCAP (Thermo Fisher Scientific; Phadia AB, Uppsala, Sweden) according to the manufacturer's recommendations. sIgE values ≥ 0.35 kU/L were considered positive, and for lower values a value of two-thirds of the detection limit (0.23 kU/L) was assigned. For data analysis, sIgE values greater than 100 kU/L were replaced by 110 kU/L. The measuring range of tIgE for an undiluted sample is 2–5000 kU/L.

2.3 Rye Flour Challenge Test

Bronchial challenge tests were performed with nebulized aqueous rye flour solutions in 24 cases, with native rye flour simulating the situation at the workplace in 63 cases, and nasal challenges in 8 cases. A nasal challenge test was considered positive if nasal symptoms were followed by a decrease of nasal flow by at least 40% from baseline. Allergen-induced airway responsiveness was measured by body plethysmography. A positive test result was assumed if specific airway resistance (sRaw) doubled and increased to ≥ 2.0 kPa's, or if the fall in forced expired volume in one second (FEV₁) was $\geq 20\%$.

2.4 Skin Prick Test

A panel of common inhalation allergens, including grass pollen, birch pollen, house dust mite (*Dermatophagoides pteronyssinus*), and cat dander (Allergopharma; Reinbek, Germany) were tested (double estimation). Atopy was defined as a mean wheal diameter ≥ 3 mm to at least one of these aeroallergens. Histamine (10 mg/mL) and saline were used as positive and negative controls, respectively.

2.5 Statistical Analysis

Concentrations of rye flour-sIgE, tIgE, and the sIgE/tIgE ratio in bakers with a positive or a negative challenge test were compared with the Mann-Whitney U test. Receiver operating characteristic (ROC) plots are one possible graphical presentation for describing and comparing diagnostic tests. The area under the curve (AUC) is a global measure of the diagnostic performance of a test and ranges from 0 to 1. ROC plots were constructed using the outcome of challenge test as gold-standard. AUCs were calculated to compare the results for rye flour-sIgE alone, tIgE, and the sIgE/tIgE ratio. For further evaluation, optimal cut-off levels that lead to the maximum Youden Index (sensitivity + specificity - 1) were calculated. Using these optimal cut-offs, sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were calculated. Correlation between variables was assessed with Spearman's correlation coefficient (r). Comparisons were considered significant at $p < 0.05$. Calculations were performed using a commercial GraphPad Prism v7.03 statistical package (GraphPad Software; La Jolla, CA).

3 Results

Sixty-three (66%) of the 95 bakers challenged with rye flour showed a positive test result. The rye flour-sIgE concentration in all bakers ranged from <0.35 to >100 kU/L (median of 1.31 kU/L), tIgE from 2.3 to 4161 kU/L (median: 98.5 kU/L), and the sIgE/tIgE ratio was between 0.003 and 19.1% (median of 2.3%). The tIgE and sIgE concentrations and the sIgE/tIgE ratio were significantly higher in bakers with a positive challenge test than in those with a negative one (Fig. 1). Overall, sIgE correlated strongly with the tIgE concentration of ($r = 0.688$; $p < 0.0001$).

The AUC of the ROC plots was 0.83 for rye flour-sIgE, 0.79 for tIgE, and 0.64 for the sIgE/tIgE ratio (Fig. 2). Sensitivity, specificity, PPV, and NPV at the optimal cut-off levels (sIgE >2.4 kU/L, tIgE

>51.8 kU/L, and sIgE/tIgE $>1.8\%$) are shown in Table 1. The maximum Youden Index was similar for sIgE and tIgE (0.556 and 0.561, respectively), while it was only half as high for sIgE/tIgE (0.276). Whereas tIgE >51.8 kU/L was a sensitive predictor of the challenge result (sensitivity 87%, NPV 75%), rye flour-sIgE was an excellent and specific predictor of a positive challenge result (rye flour-sIgE >2.4 kU/L: specificity 94%, PPV 95%).

According to skin prick testing with ubiquitous allergens, 44 bakers showed a positive reaction to at least one of the allergens and were defined as atopics. The remaining 51 bakers were defined to be non-atopic. While the concentrations of rye flour-sIgE and tIgE were significantly higher in atopic bakers, no significant difference was obtained for the sIgE/tIgE ratio (Table 2). ROC analysis stratified according to atopy is shown in Fig. 3. While in the group of non-atopic bakers, the AUC for rye flour-sIgE and tIgE were approximately 50% higher than that for their ratio, in the group of atopic bakers all three AUC were in a similar range.

4 Discussion

According to Kleine-Tebbe (2012), IgE results of symptomatic subjects should generally be checked for sIgE to tIgE ratio. In the present study we evaluated the ratio of rye flour-sIgE to tIgE in a group of 95 bakers with work-related allergic symptoms to determine whether the ratio could be superior to sIgE alone in the diagnosis of baker's asthma. As shown by the AUC values of ROC curves, the sIgE/tIgE ratio failed to improve the prediction of the outcome of a challenge test with rye flour. This finding was also true for the stratified analysis of both atopic and non-atopic bakers.

Gupta et al. (2014) have reported that the sIgE/tIgE ratio is significantly more accurate than sIgE alone in predicting the oral challenge outcome in children with suspected food allergy (AUC 0.69 vs. 0.55; $p = 0.03$). In contrast, in another study this ratio has not been as efficacious as sIgE alone for the diagnosis of symptomatic food allergy in children and therefore judged of no real benefit

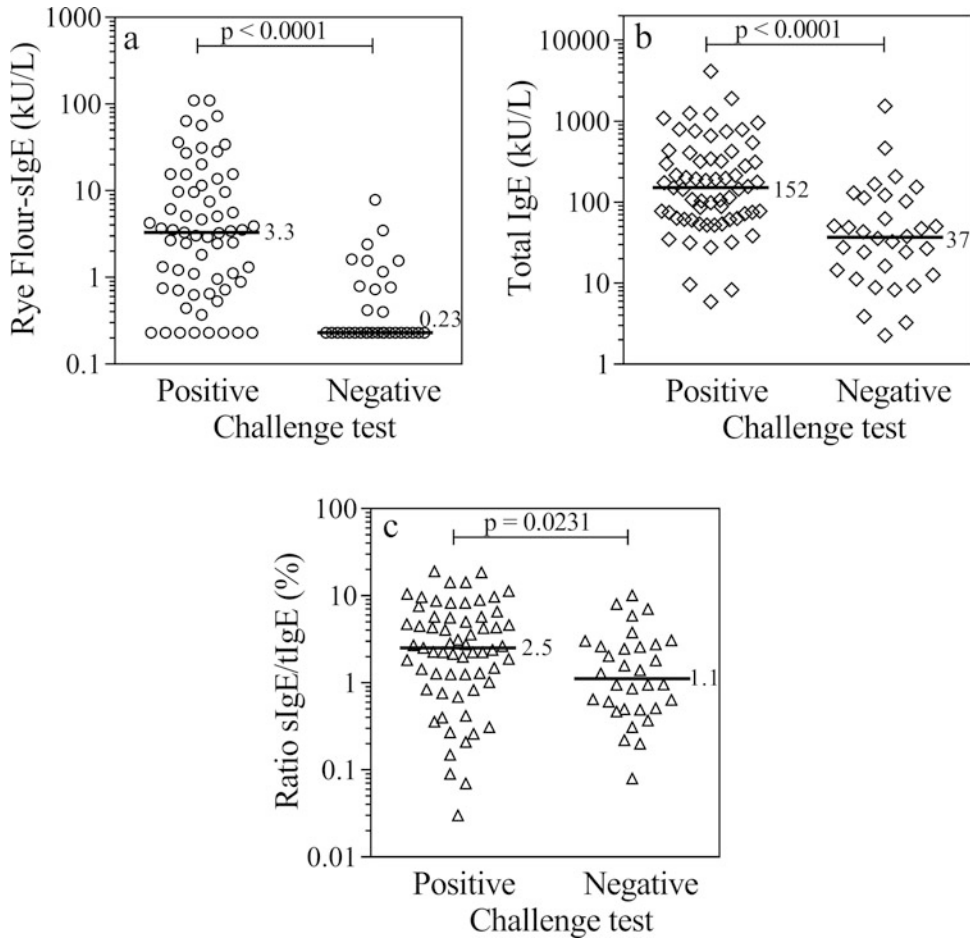


Fig. 1 Comparison of concentrations of rye flour-sIgE (a), total IgE (tIgE) (b), and the ratio of sIgE/tIgE (c) between bakers with a positive challenge test (n = 63) and

those with a negative challenge test (n = 32) to rye flour. Horizontal lines in each panel represent medians

(Mehl et al. 2005). In the latter study, differences in the AUCs of ROC curves depended on the allergen. The respective AUCs were 0.79 (ratio) vs. 0.76 (sIgE alone) for cow's milk, 0.86 vs. 0.88 for hen's egg, 0.66 vs. 0.66 for wheat, and 0.58 vs. 0.64 for soy. For the allergens above outlined, the maximal sIgE/tIgE ratio amounted to 91.2% (median: 0.3%) for cow's milk, 69.4% (median: 1.7%) for hen's egg, 70.7% (median: 0%) for wheat, and 15.0% (median: 0%) for soy. From this ranking, one could suppose that the utility of the ratio in predicting the outcome of a challenge test increases with the height of the ratio. This could be an explanation for the low diagnostic

performance of the rye flour-sIgE/tIgE ratio in the present study (AUC 0.64 (ratio) vs. 0.83 (sIgE alone)) because the maximum ratio was only 19.1% (median: 2.3%).

However, the afore-mentioned studies were performed in children suffering from food allergy, while this study concerns adult bakers with respiratory work-related symptoms subjected to inhalation exposure to rye flour. In this connection, it is noteworthy that the level of tIgE is age-dependent. In a study including 603 children, aged between 1 and 9 years, with suspected food allergy a significant positive correlation between the total serum IgE level and

patients' age has been observed (Horimukai et al. 2015). On the other hand, a large longitudinal study spanning more than a decade in 6371 young and middle-aged adults (20–44 years) has shown that tIgE decreases in elderly subjects while there was no significant change in the prevalence of sensitization to at least one environmental allergen over the study period (Jarvis et al. 2005).

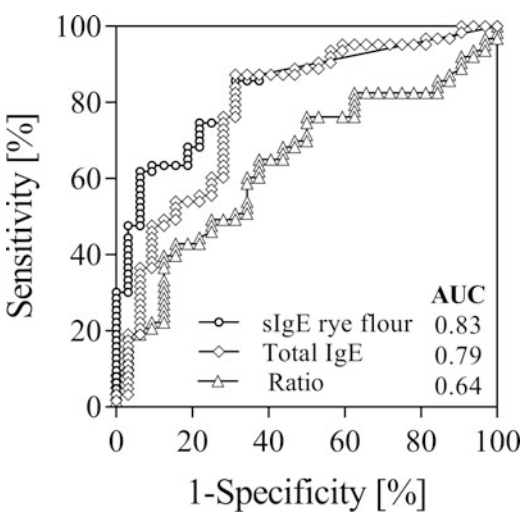


Fig. 2 Receiver operating curves (ROC) for rye flour-sIgE, total IgE (tIgE), and the ratio of sIgE/tIgE of 95 symptomatic bakers with the challenge test as gold-standard. The area under curve (AUC) is a global measure for the test's accuracy that ranges between 0 and 1

One study enrolling 171 adult β -lactam allergic patients and 122 controls shows that the sIgE/tIgE ratio could improve the diagnosis of β -lactam allergy (Vultaggio et al. 2015). Due to the fact that the authors compared the diagnostic performance of sIgE to at least one β -lactam hapten with the ratio that was calculated by making the sum of sIgE to all four measured β -lactam haptens divided by tIgE, a comparison of the usefulness of sIgE alone and the ratio, like we did it in the present study, was not possible. In addition, a skin prick test with β -lactams rather than a challenge test was used as gold-standard in that previous study. Nonetheless, the authors hypothesize that a high sIgE/tIgE ratio increases the probability to have two neighboring sIgE molecules on the basophil/mast cell surface membrane, which could lead to an easier cross-linking of the high-affinity IgE receptor with cell activation that follows.

An additional finding of the present study was that the level of tIgE resulted in a much better prediction of the outcome of a challenge test with rye flour than the ratio of rye flour-sIgE/tIgE (AUC: 0.79 vs. 0.64), although it could be expected due to a high correlation between rye flour-sIgE and tIgE. To the best of our knowledge, so far no study has evaluated the predictive value of tIgE for the outcome of an inhalation allergen challenge, but it is known that high levels of tIgE strongly increase the probability of sensitization,

Table 1 Sensitivity, specificity, positive (PPV) and negative predictive value (NPV) of rye flour-specific IgE (sIgE), total IgE (tIgE), and the sIgE/tIgE ratio, based on the gold-standard challenge test

	Cut-off	Maximum Youden Index	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
sIgE (kU/L)	> 2.4	0.556	61.9	93.8	95.1	57.0
tIgE (kU/L)	> 51.8	0.561	87.3	68.8	84.4	74.5
sIgE/tIgE (%)	> 1.8	0.276	65.1	62.5	77.1	49.1

Evaluation was performed using the cut-offs which were obtained by the maximum Youden Index

Table 2 Comparison of concentrations of rye flour-specific IgE (sIgE), total IgE (tIgE), and the sIgE/tIgE ratio between atopic and non-atopic bakers

	Atopics (n = 44) median (range)	Non-atopics (n = 51) median (range)	p-value
sIgE (kU/L)	2.75 (0.23–110)	0.72 (0.23–57.1)	0.007
tIgE (kU/L)	134 (5.91–4161)	72 (2.28–1536)	0.036
sIgE/tIgE (%)	2.25 (0.03–18.60)	2.04 (0.07–19.10)	0.401

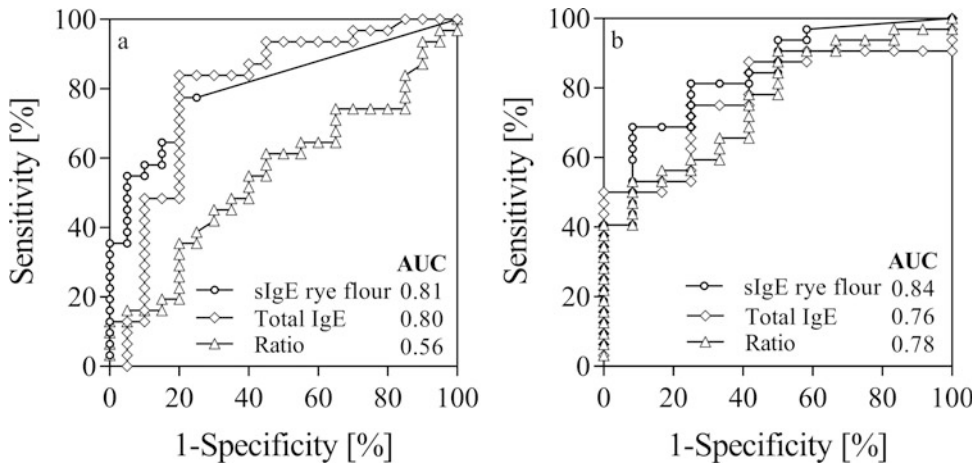


Fig. 3 Receiver operating curves (ROC) stratified according to atopy status. Rye flour-sIgE, total IgE (tIgE), and the sIgE/tIgE ratio of 44 atopic (a) and

51 non-atopic (b) bakers were evaluated with the challenge test as gold-standard; AUC, area under curve

especially in young individuals (Kerkhof et al. 2003) and also are predictive of asthma (Ahmad Al Obaidi et al. 2008). However, tIgE should not be used as definite evidence for an allergic disease since its level is influenced by numerous factors, especially age (Chang et al. 2015).

In conclusion, rye flour-sIgE is superior to the ratio of rye flour-sIgE to tIgE, independent from atopy status, as a diagnostic test to distinguish between bakers with and without occupational asthma to rye flour. Overall, the accuracy of the ratio, as a diagnostic test, was not satisfactory in the present study. Whereas tIgE resulted in a high negative predicted value and was more suitable to rule out a positive result of a challenge test, it could be confirmed that a high concentration of rye flour-sIgE in the sera of bakers suffering from work-related symptoms is a good predictor of a positive challenge result. Thus, specific challenges with flours may be omitted in strongly sensitized bakers (van Kampen et al. 2008).

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Conflicts of Interest The authors declare no conflicts of interest in relation to this article.

References

- Ahmad Al Obaidi AH, Mohamed Al Samarai AG, Yahya Al Samarai AK, Al Janabi JM (2008) The predictive value of IgE as biomarker in asthma. *J Asthma* 45:654–663
- Buslau A, Voss S, Herrmann E, Schubert R, Zielen S, Schulze J (2014) Can we predict allergen-induced asthma in patients with allergic rhinitis? *Clin Exp Allergy* 44:1494–1502
- Chang M-L, Cui C, Liu Y-H, Pei L-C, Shao B (2015) Analysis of total immunoglobulin E and specific immunoglobulin E of 3,721 patients with allergic disease. *Biomed Rep* 3:573–577
- DGUV (2015) Deutsche Gesetzliche Unfallversicherung Statistics, Sankt Augustin (in German)
- Douglas TA, Kusel M, Pascoe EM, Loh RK, Holt PG, Sly PD (2007) Predictors of response to bronchial allergen challenge in 5- to 6-year-old atopic children. *Allergy* 62:401–407
- Fernández C, Cárdenas R, Martín D, Garcimartín M, Romero S, de La Cámara AG, Vives R (2007) Analysis of skin testing and serum-specific immunoglobulin E to predict airway reactivity to cat allergens. *Clin Exp Allergy* 37:391–399
- Grabenhenrich L, Lange L, Härtl M, Kalb B, Ziegert M, Finger A, Harandi N, Schlags R, Gappa M, Puzzo L, Stephan V, Heigle T, Büsing S, Ott H, Niggemann B, Beyer K (2016) The component-specific to total IgE ratios do not improve peanut and hazelnut allergy diagnoses. *J Allergy Clin Immunol* 137:1751–1760
- Gupta RS, Lau CH, Hamilton RG, Donnell A, Newhall KK (2014) Predicting outcomes of oral food

- challenges by using the allergen-specific IgE-total IgE ratio. *J Allergy Clin Immunol Pract* 2:300–305
- Horimukai K, Hayashi K, Tsumura Y, Nomura I, Narita M, Ohya Y, Saito H, Matsumoto K (2015) Total serum IgE level influences oral food challenge tests for IgE-mediated food allergies. *Allergy* 70:334–337
- Jarvis D, Luczynska C, Chinn S, Potts J, Sunyer J, Janson C, Svanes C, Künzli N, Leynaert B, Heinrich J, Kerkhof M, Ackermann-Lieblich U, Antó JM, Cerveri I, de Marco R, Gislason T, Neukirch F, Vermeire P, Wjst M, Burney P (2005) Change in prevalence of IgE sensitization and mean total IgE with age and cohort. *J Allergy Clin Immunol* 116:675–682
- Kerkhof M, Dubois AE, Postma DS, Schouten JP, de Monchy JG (2003) Role and interpretation of total serum IgE measurements in the diagnosis of allergic airway disease in adults. *Allergy* 58:905–911
- Kleine-Tebbe J (2012) Old questions and novel clues. Complexity of IgE repertoires. *Clin Exp Allergy* 42:1142–1145
- Mehl A, Verstege A, Staden U, Kulig M, Nocon M, Beyer K, Niggemann B (2005) Utility of the ratio of food-specific IgE/total IgE in predicting symptomatic food allergy in children. *Allergy* 60:1034–1039
- Muñoz X, Cruz MJ, Bustamante V, Lopez-Campos JL, Barreiro E (2014) Work-related asthma. Diagnosis and prognosis of immunological occupational asthma and work-exacerbated asthma. *J Investig Allergol Clin Immunol* 24:396–405
- Soares-Weiser K, Takwoingi Y, Panesar SS, Muraro A, Werfel T, Hoffmann-Sommergruber K, Roberts G, Halken S, Poulsen L, van Ree R, Vlieg-Boerstra BJ, Sheikh A (2014) The diagnosis of food allergy. A systematic review and meta-analysis. *Allergy* 69:76–86
- van Kampen V, Rabstein S, Sander I, Merget R, Brüning T, Broding HC, Keller C, Müsken H, Overlack A, Schultze-Werninghaus G, Walusiak J, Raulf-Heimsoth M (2008) Prediction of challenge test results by flour-specific IgE and skin prick test in symptomatic bakers. *Allergy* 63:897–902
- Vandenplas O, Froidure A, Meurer U, Rihs H-P, Riffart C, Soetaert S, Jamart J, Pilette C, Raulf M (2016) The role of allergen components for the diagnosis of latex-induced occupational asthma. *Allergy* 71:840–849
- Vandenplas O, Suojalehto H, Cullinan P (2017) Diagnosing occupational asthma. *Clin Exp Allergy* 47:6–18
- Vultaggio A, Virgili G, Gaeta F, Romano A, Maggi E, Matucci A (2015) High serum β -lactams specific/total IgE ratio is associated with immediate reactions to β -lactams antibiotics. *PLoS One* 10(4):e0121857